Fact Sheet for Health Care Providers: Interpreting Zika MAC-ELISA Results

June 29, 2016

Dear Health Care Provider:

The U.S Food and Drug Administration (FDA) has issued an Emergency Use Authorization (EUA) to authorize the use of the Centers for Disease Control and Prevention's (CDC) Zika IgM antibody capture ELISA (Zika MAC-ELISA). This assay provides *in vitro* qualitative detection of human IgM antibodies to Zika virus. It is intended for use in sera or cerebrospinal fluid (CSF) when submitted with a patient-matched serum sample from individuals meeting CDC Zika clinical and epidemiological criteria for testing (http://www.cdc.gov/zika/hc-providers/index.html) in qualified laboratories designated by the CDC. The test is intended for use as part of CDC's algorithm for Zika testing.

FDA issued this EUA based on data submitted by CDC to FDA, and on the U.S. Secretary of Health and Human Services' (HHS) declaration that circumstances exist to justify the emergency use of *in vitro* diagnostic tests for the detection of Zika virus and Zika virus infection. This EUA will terminate when the HHS Secretary's declaration terminates, unless FDA revokes it sooner.

The information in this Fact Sheet is to inform you of the significant known and potential risks and benefits of the emergency use of the Zika MAC-ELISA. For more information on this EUA, please see FDA's website at http://www.fda.gov/MedicalDevices/Safety/EmergencySituations/ucm161496.htm.

Why is this test needed at this time?

As of June 22, 2016, active Zika virus transmission is occurring in 39 countries and territories in the Americas, 8 countries and territories in Oceania/Pacific Islands, and 1 country in Africa (http://www.cdc.gov/zika/geo/active-countries.html). Among cases identified in 2015-16, Zika virus transmission has occurred primarily through the bite of infected *Aedes* species mosquitoes. Zika virus can also be transmitted from mother to fetus during pregnancy and through sexual transmission from infected males to their sexual partners.

As of June 22, 2016, over 819 confirmed cases of Zika virus infection have been identified in the continental United States. All cases were in persons with either a recent travel history to areas with ongoing transmission or an epidemiologic link with an individual with such travel history (i.e., through maternal-fetal or sexual transmission). Public health officials have determined that Zika virus poses a potential public health emergency.

At this time, there are no FDA approved/cleared tests available that can detect Zika virus in clinical specimens in the United States. Therefore, CDC has developed this test to detect evidence of Zika virus infections in human sera and CSF. Current information on Zika virus infection for health care providers, including case definitions, is available at

http://www.cdc.gov/zika/hc-providers/index.html. All information and guidelines, including those on Zika virus laboratory testing, may change as more data is gathered on this virus. Please check CDC's Zika Virus website regularly for the most current information (http://www.cdc.gov/zika/index.html).

If Zika virus infection is suspected based on current clinical and/or epidemiological criteria recommended by public health authorities, the Zika MAC-ELISA may be ordered. As chikungunya virus infection and dengue virus infection can have early symptoms resembling those of Zika virus, testing should be considered for chikungunya and dengue. Please contact your state or local health department to facilitate testing.

The results should be used in conjunction with clinical signs and symptoms, epidemiological information, and travel history to diagnose recent Zika virus infection. This test is authorized for use with serum and with CSF (when submitted with a patient-matched serum sample).

As of February 20, 2016, serum is the primary diagnostic specimen and should be the priority specimen for collection and testing. Specimens should be collected with appropriate infection control precautions and according to the manufacturer's instructions for the specimen collection device. Sera should be collected in serum separator tubes and centrifuged after collection to reduce the likelihood of hemolysis. Please refer to manufacturer's instructions for serum tube processing. Additional guidance for collection of body fluid specimens for Zika diagnostic testing may be found at: http://www.cdc.gov/zika/hc-providers/body-fluids-collection-submission.html.

What are the symptoms of Zika virus infection?

Many people with Zika virus infection are asymptomatic. Symptomatic patients typically experience a mild illness characterized by fever, rash, joint pain, or conjunctivitis. Clinical illness is usually self-limited and lasts a week or less. Clinical illness recognition can be complicated in that not all symptomatic patients report all of these symptoms, and Zika manifestations overlap significantly with those seen in other viral infections. Although the exact incubation period is yet to be determined, it is considered to be about 3 days to 2 weeks.

Based on a review of available evidence, CDC has concluded that Zika virus infection in pregnancy is a cause of microcephaly (a birth defect characterized by small head size and impaired cranial and neural development in fetuses and infants) and other serious abnormalities of the brain. In addition, it has been linked to central nervous system injury, placental insufficiency, fetal growth restriction, and fetal loss, eye abnormalities, and hearing impairment (references 1-2).

Limited information is available currently about the spectrum of defects caused by prenatal Zika virus infection, the relative and absolute risks of adverse outcomes among fetuses whose mothers were infected at different times during pregnancy, and factors that might affect a woman's risk of adverse pregnancy or birth outcomes.

It is also important to note that Zika virus infection is not the sole suspected cause of microcephaly in fetuses and infants.

There are also reports of Guillain-Barre syndrome associated with Zika virus infection.

When should the Zika MAC-ELISA be performed?

Anti-Zika IgM is typically detectable starting soon after onset of symptoms and is reliably detectable for approximately 12 weeks following infection. The Zika MAC-ELISA test should be performed according to the CDC-issued algorithm available at http://www.cdc.gov/zika/state-labs/index.html.

What does it mean if the specimen tests positive in the Zika MAC-ELISA?

A positive test result for Zika virus from the Zika MAC-ELISA indicates that anti-Zika IgM antibodies were detected in the sera or CSF of the patient. Confirmation of Zika MAC-ELISA positive or equivocal results requires additional testing by CDC or by qualified laboratories designated by CDC and in consultation with CDC, using the CDC-issued algorithm found at: http://www.cdc.gov/zika/state-labs/index.html.

False positive serological results are possible (see next paragraph). Laboratory test results should always be considered in the context of clinical observations and epidemiologic information in making a final diagnosis and patient management decisions. Any positive test result for Zika virus infection, including Zika MAC-ELISA positive results, should be reported to your local or state health department. In the United States and its territories, Zika virus disease and congenital Zika virus infection are nationally notifiable diseases. For guidelines on Zika virus, please refer to http://www.cdc.gov/zika/hc-providers/index.html.

Positive and equivocal Zika MAC-ELISA results are not definitive for diagnosis of Zika virus infection. False positive results may occur in some patients with recent, closely-related flavivirus infections, such as dengue infections. In patients who have received yellow fever or Japanese encephalitis vaccination, cross-reactive antibodies in both the IgM and neutralizing antibody assays may make it difficult to identify which flavivirus is causing the patient's current illness. It is possible that the Zika MAC-ELISA may generate positive results in patients with a history of non-Zika flavivirus infections. In the event of a false positive result, risks to patients could include any or all of the following: the impaired ability to detect and receive appropriate medical care for the true infection causing the symptoms, an unnecessary increase in the monitoring of a woman's pregnancy, or other unintended adverse effects.

It should be emphasized that the identification of possible Zika virus infection in a pregnant woman does not provide any definitive information about the state of health of the fetus. Many questions remain about the association between Zika virus infection in a mother and the impact to the fetus, and the impact of factors such as timing, likelihood, and relevance of symptomatic versus asymptomatic infection. Detection of Zika virus infection in the mother does not mean there is definite harm to the fetus.

What does it mean if the specimen tests negative in the Zika MAC-ELISA?

A negative Zika MAC-ELISA result does not rule out Zika virus infection, particularly if testing is conducted soon after onset of symptoms (before IgM levels are expected to become detectable) or more than 12 weeks after the infection is thought to have occurred (as IgM levels are expected to drop). As with any test, providers must consider the patient's likelihood of exposure and the possibility of false laboratory results when making treatment or other patient management decisions. The possibility of a false negative result should especially be considered if the patient's recent exposures or clinical presentation are consistent with Zika virus infection and diagnostic tests for other causes of illness are negative. Conversely, a negative result in an asymptomatic patient with a lower likelihood of exposure (e.g., a short term traveler to an affected area) may suggest the patient is not infected.

Please refer to CDC guidance for Health Care Providers Caring for Pregnant Women and Women of Reproductive Age with Possible Zika Virus Exposure:

http://www.cdc.gov/zika/hc-providers/clinical-guidance.html

Reporting Adverse Events

You should report adverse events, including problems with test performance or results, to MedWatch at http://www.fda.gov/medwatch, by submitting a MedWatch Form 3500 (available at http://www.fda.gov/medwatch/safety/FDA-3500_fillable.pdf) or by calling 1-800-FDA-1088.

Pregnant patients should receive the Fact Sheet for Pregnant Women: Understanding Results from the Zika MAC-ELISA. All other patients should receive the Fact Sheet for Patients: Understanding Results from the Zika MAC-ELISA.

Contact Information for the Manufacturer:
CDC Emergency Operations Center (EOC)
1600 Clifton Road
Atlanta, Georgia, USA, 30329
Office phone: **CDC EOC (770-488-7100)**

Any significant new findings that negatively impact the performance of the test and that are observed during the course of the emergency use of the Zika-MAC ELISA will be made available at http://www.cdc.gov/zika/index.html.

References

1) Rasmussen S.A., Jamieson D.J., Honein M.A., and Petersen L.R. Zika Virus Birth Defects—Reviewing the Evidence for Causality. *NEJM* (April 12, 2016). DOI: 10.1056/NEJMsr1604338.

2) CDC Website. http://www.cdc.gov/zika.

Fact Sheet for Patients:
Understanding Results from the Zika MAC-ELISA

June 29, 2016

Dear Patient:

If you are pregnant, please ask your doctor for the Fact Sheet for Pregnant Women: Understanding Results from the Zika MAC-ELISA.

You are being given this Fact Sheet because your blood or cerebrospinal fluid (CSF) has been tested for evidence of Zika virus infection. This testing was done because your health care provider believes you may have been exposed to the virus. The Zika MAC-ELISA is a test to help determine if you have recently been infected with Zika virus.

This Fact Sheet contains information to help you understand the risks and benefits of using the Zika MAC-ELISA. If possible, you may want to discuss with your health care provider the risks and benefits described in this Fact Sheet and any additional questions you may have.

What is Zika virus Infection?

Zika virus infection is caused by the Zika virus, which is most often spread to people through mosquito bites. A woman infected with Zika virus during pregnancy can pass the virus to her developing baby. Zika virus can also be passed by an infected man to his partner during sex. Since 2015, a large number of people infected with Zika virus have been reported in many South and Central American and Caribbean countries.

Most people who are infected with Zika virus do not have any symptoms. Those that do usually have mild illness with symptoms that may include fever, rash, joint pain, or redness of the eyes. These symptoms often resolve on their own within a week.

Infection with Zika virus during pregnancy can cause microcephaly (where the baby's head is smaller than expected, a sign of incomplete brain development) and other severe brain defects. However, detection of Zika virus infection in the mother does not mean there is definite harm to the developing baby. Some women who had Zika virus infection during pregnancy have delivered apparently healthy babies. Women who are infected with Zika virus while pregnant should be monitored more closely by their health care providers throughout their pregnancy.

There have also been reports of a possible link between Zika virus infection and an illness that can cause temporary paralysis (Guillain-Barré syndrome).

What is the Zika MAC-ELISA?

The Zika MAC-ELISA is a laboratory test designed to detect proteins the human body makes to fight a Zika virus infection. These proteins, called antibodies, appear in the blood starting soon after the start of illness and last for up to 12 weeks. In some people, they are present

for longer than 12 weeks. If the Zika MAC-ELISA detects these antibodies, the test is positive. If the Zika MAC-ELISA does not detect these antibodies, the test is negative.

The Zika MAC-ELISA has not been cleared or approved by the U.S. Food and Drug Administration (FDA). However, FDA has authorized the emergency use of this test under an Emergency Use Authorization (EUA).

Why was my sample tested using the Zika MAC-ELISA?
Your blood or CSF sample(s) were tested because you have symptoms that resemble Zika virus infection, because you live in or have traveled recently to a place where Zika virus infection is known to occur, and/or because you have another possible exposure to Zika virus (e.g., sexual transmission). The sample(s) collected from you was tested using the Zika MAC-ELISA to help find out whether you may have been recently infected with Zika virus. The test results, along with other information, could help your health care provider make decisions about how to take care of you and may help to limit the spread of Zika virus in your community.

What are the known risks and benefits of the CDC Zika MAC-ELISA?
Besides possible discomfort or other complications that can happen when your specimen is collected, there is a risk that the test result will be incorrect (see next paragraphs for more information). The benefit of having this test is that the results of this test, along with other information, can help your health care provider make decisions about how to take care of you. Also, knowing your test results may help keep you from giving Zika virus to others (e.g., by allowing you to take measures to avoid sexual transmission of the virus to someone else).

If this test is positive for Zika virus, does it mean that I have Zika virus infection?
If you have a positive result with the Zika MAC-ELISA, it is likely that you recently were infected with the Zika virus. There is a chance that this test can give a positive result that is wrong; this is called a “false positive” result. There are some other very closely related viruses (such as dengue virus) that can cause the human body to produce antibodies that may cause the test to be positive.

If your result from this test is positive or equivocal (unclear), your healthcare provider or health department will determine if your results should be evaluated with additional testing and/or with testing from other samples that may have been collected from you. It is important that you work with your health care provider or health department to help you understand the next steps you should take. Information about steps to take if you are diagnosed with Zika virus infection is available at http://www.cdc.gov/zika/index.html.

If you are male and have a positive test result for Zika virus, you should either use a condom the right way every time or not have sex with your partner during pregnancy. If you are female and have a positive test result and you are considering becoming pregnant, then you should discuss the risks with your health care provider.

More information about Zika virus infection, including how to prevent sexual transmission of Zika virus and information for women and their partners who are thinking about pregnancy, is available at http://www.cdc.gov/zika/index.html.

If this test is negative, does it mean that I do not have Zika virus infection?
Even if you have a negative test, you may have been infected with Zika virus. If your sample was collected just after you became ill, it is possible that your body had not yet had enough time to make antibodies for the test to measure. If the sample was collected more than 12 weeks after your illness, it is possible that your body has already fought off the virus and the amount of antibodies is so low that they cannot be measured. Your health care provider will help you to interpret your test results and work with you to continue to monitor your health.

What is an Emergency Use Authorization (EUA)?
An EUA is a tool that FDA can use to allow the use of certain medical products for emergencies based on scientific data. The U.S. Secretary of Health and Human Services (HHS) has declared that circumstances exist to allow the emergency use of authorized diagnostic tests for Zika virus infection, such as the Zika MAC-ELISA, under an EUA.

At this time, there are no FDA approved/cleared alternative tests available that detect Zika virus infection. FDA has authorized the emergency use of the Zika MAC-ELISA to test for antibodies to Zika virus in blood and CSF. Use of this test is authorized only for the duration of the potential emergency, unless it is terminated or revoked by FDA sooner.

How can I learn more?
Information about Zika virus and any significant new findings that negatively impact the performance of the test and that are observed during the course of the emergency use of the Zika MAC-ELISA will be made available at the CDC website:
http://www.cdc.gov/zika/index.html

Please also contact your health care provider if you have any questions.

Fact Sheet for Pregnant Women:
Understanding Results from the Zika MAC-ELISA

June 29, 2016

Dear Madam:

You are being given this Fact Sheet because your blood or cerebrospinal fluid (CSF) has been tested for evidence of Zika virus infection. This testing was done because your health care provider believes you may have been exposed to the virus. The Zika MAC-ELISA is a test to help determine if you have recently been infected with Zika virus.

This Fact Sheet contains information to help you understand the risks and benefits of using the Zika MAC-ELISA. If possible, you may want to discuss with your health care provider the risks and benefits described in this Fact Sheet and any additional questions you may have.

What is Zika virus Infection?

Zika virus infection is caused by the Zika virus, which is most often spread to people through mosquito bites. A woman infected with Zika virus during pregnancy can pass the virus to her developing baby. Zika virus can also be passed by an infected man to his partner during sex. Since 2015, a large number of people infected with Zika virus have been reported in many South and Central American and Caribbean countries.

Most people who are infected with Zika virus do not have any symptoms. Those that do usually have mild illness with symptoms that may include fever, rash, joint pain, or redness of the eyes. These symptoms often resolve on their own within a week.

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The Zika MAC-ELISA has not been cleared or approved by the U.S. Food and Drug Administration (FDA). However, FDA has authorized the emergency use of this test under an Emergency Use Authorization (EUA).

Why was my sample tested using the Zika MAC-ELISA?
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What are the known risks and benefits of the CDC Zika MAC-ELISA?
Besides possible discomfort or other complications that can happen when your specimen is collected, there is still a risk that the test result will be incorrect (see next paragraphs for more information). The benefit of having this test is that the results of this test, along with other information, can help your health care provider make decisions about how to take care of you and your developing baby.

If this test is positive for Zika virus, does it mean that I have Zika virus infection?
If you have a positive result with the Zika MAC-ELISA, it is likely that you recently were infected with the Zika virus. There is a chance that this test can give a positive result that is wrong; this is called a "false positive" result. There are some other very closely related viruses (such as dengue virus) that can cause the human body to produce antibodies that may cause the test to be positive.

If your result from this test is positive or equivocal (unclear), your health care provider or health department will determine if your results should be evaluated with additional testing and/or with testing from other samples that may have been collected from you. It is important that you work with your health care provider or health department to help you understand the next steps you should take. They will also work closely with you to monitor the health and development of your baby.

If this test is positive for Zika virus, does it mean that my child will have a birth defect?
No, not necessarily. Studies have reported that some, but not all, babies born to women with positive Zika test results during pregnancy were born with microcephaly and other problems. At this time, we do not know how often a baby will have microcephaly or other problems if a woman is infected with Zika while she is pregnant. Your doctor or other health care provider will watch your pregnancy more closely if you have a positive Zika virus test. This test can also give positive results when the patient has had an infection with a virus other than Zika virus (see above). The results of this test are not conclusive - a positive test result for Zika virus infection during pregnancy signals to your doctor or other healthcare provider to watch your pregnancy more closely, meaning he or she might do more ultrasounds or other tests to check the growth and development of your fetus and check for any signs of Zika virus infection.

If this test is negative, does it mean that I do not have Zika virus infection?
Even if you have a negative test, you may have been infected with Zika virus. If your sample was collected just after you became ill, it is possible that your body had not yet had enough time to make antibodies for the test to measure. If the sample was collected more than 12 weeks after your illness, it is possible that your body has already fought off the virus and the amount of antibodies is so low that they cannot be measured. Your health care provider will help you to interpret your test results and work with you to continue to monitor your health and the health of your baby.

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